CYP2C9 and CYP3A4 enzymes and P-gp and BCRP efflux transporters. In addition, the bioprinted intestinal tissues respond to known toxicants indomethacin and TNFα with reduced barrier function, increased cytotoxicity, and changes in gene expression and cell morphology. The fully human primary cell-derived tissue combined with the reproducibility of the bioprinting platform and compatibility with standard assay approaches make this system a novel and practical in vitro tool for ADME/Tox applications across drug development.

SUMMARY OF THE INVENTION

[0011] The present invention provides intestinal tissue models that offer advantages over existing in vitro assay systems by providing an intestinal epithelial cell layer on top of a layer of intestinal interstitial tissue comprising myofibroblasts and, optionally, other key cell types such as myeloid immune cells, smooth muscle cells, endothelial cells and neurons. These intestinal tissue models allow one to see the impact of treatments in a more holistic system. And, the intestinal tissue models support epithelial morphology and function for an extended period of time in culture, thus enabling chronic studies of treatments. Because of their multi-cellularity and architecture, the intestinal tissue constructs provide a unique system to study multi-faceted processes including secretion, transport, cell-cell interactions and pathogenic processes, including inflammation and cancer.

[0012] The intestinal tissue models are valuable alternatives to animal models in the pharmaceutical industry for ADME-TOX applications in the lead optimization stage of drug development as well as disease modeling across all phases of drug discovery. In one embodiment, the intestinal tissue models described herein incorporate both epithelium and lamina propria to approximate the intestinal mucosa. In another embodiment, the tissue constructs comprise primary human intestinal epithelial cells in an epithelial compartment supported by primary human myofibroblasts in an interstitial compartment. The complexity of the model is optionally increased by incorporating additional specialized cell types, for example, enteroendocrine cells, into the epithelial layer to model endocrine function while goblet cells can be added to model mucosal barrier function. Additional complexity is achieved by the addition of immune cells or a submucosal compartment comprising endothelial cells and smooth muscle cells. An advantage of disclosed intestinal tissue constructs is that they are more physiologically relevant compared to tissue constructs having two-dimensional environment. The multi-cellularity and architecture of the tissues provide a unique opportunity to study complex multi-faceted processes of cells in a three-dimensional conformation including secretion, transport, cell-cell interactions and pathogenic processes. Through these interactions, three-dimensional tissues differentiate in a different manner than cells cultured in a two-dimensional monolayer, activating new signaling pathways and extracellular matrix interactions.

[0013] The intestinal tissue models described herein provide an opportunity to accurately study how compounds affect the intestinal tissue as well as to model pathogenic processes in the intestine. The intestinal tissue models disclosed herein are useful for predicting toxicity of pharmaceutical compounds earlier in the drug development process. By incorporating both diseased (ex: inflamed or tumor tissue) and normal cell compartments into the same tissue, the impact of therapeutic agents on both healthy and diseased tissue can be assessed in the same tissue system. Intestinal tissue constructs comprising primary cells are especially useful for personalized medicine.

[0014] Unexpectedly, the intestinal tissue models described herein made with only primary epithelial cells in the layer of intestinal epithelial cells express chromogranin A, secrete glucagon-like peptide-1 (GLP-1) and mucus, manifest the formation of goblet cells and secondary structure characteristic of native intestinal tissue, tissue thickening in culture, and CYP3A4 activity. This indicates that functional enteroendrocrine cells were produced.

[0015] In one embodiment, the invention provides a three-dimensional, engineered, bioprinted, biological intestinal tissue model comprising:

[0016] (i) a layer of intestinal interstitial tissue comprising myofibroblasts; and

[0017] (ii) a layer of intestinal epithelial cells on the layer of intestinal interstitial tissue, to form the three-dimensional, engineered, biological intestinal tissue model.

[0018] In some embodiments, at least one of the layer of intestinal interstitial tissue comprises myofibroblasts and layer of intestinal epithelial cells further comprises at least one type of immune cells. In some embodiments, the immune cells are myeloid cells. In some embodiments, the myeloid cells are monocytes, macrophages, neutrophils, basophils, eosinophils, dendritic cells or megakaryocytes. In some embodiments the immune cells are lymphoid cells. In some embodiments, the immune cells are present in at least one of (a) the interstitial layer, (b) the epithelial cell layer, (c) between the interstitial layer and the epithelial cell layer, (d) on top of the epithelial cell layer, and (e) below the interstitial cell layer.

[0019] In some embodiments, the layer of intestinal epithelial cells comprises primary epithelial cells from a healthy donor. In some embodiments, the layer of intestinal epithelial cells comprises primary epithelial cells from a diseased donor. In some embodiments, the diseased donor has celiac disease, Crohn's disease, ulcerative colitis, irritable bowel syndrome, hemorrhoids, diverticulitis, inflammatory bowel disease, microscopic colitis, lymphocytic colitis, collagenous colitis, an endocrine disorder, a metabolic disorder, obesity, diabetes, dyslipidemia, intestinal or colorectal cancer.

[0020] In some embodiments, the layer of intestinal epithelial cells further comprises at least one stem cell population. In some embodiments, the at least one stem cell population is capable of differentiating. In some embodiments, the intestinal tissue model further comprises tumor (s), tumor fragment(s), tumor cells or immortalized cells. In some embodiments, the tumor(s), tumor fragment(s), tumor cells or immortalized cells are colorectal tumor(s), tumor fragment(s), tumor cells or immortalized cells. In some embodiments, the tumor(s), tumor fragment(s), tumor cells or immortalized cells are present in a layer or compartment within the intestinal tissue model.

[0021] In some embodiments, the layer of intestinal epithelial cells and layer of interstitial tissue comprises primary epithelial cells from a healthy donor. In some embodiments, the layer of intestinal epithelial cells and layer of interstitial tissue comprises primary epithelial cells from a diseased donor.